University of Illinois
Institute of Natural Resource Sustainability
William Shilts, Executive Director

ILLINOIS NATURAL HISTORY SURVEY
Brian D. Anderson Director
1816 South Oak Street
Champaign, IL 61820-6964
217-333-6830

Utility of Anuran Call-Survey Data for Estimating Occupancy, Abundance, and Reproductive Success in the Cache River Basin

Final Report

Abigail J. Maley Berkey and Christopher A. Phillips

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Restricted Access T&E Species Locations

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ILLINOIS NATURAL HISTORY SURVEY

OFFICE OF RESOURCE CONSERVATION

State of Illinois

Grant Proposal

PROJECT NUMBER: T-40-R-1

PROJECT TITLE**:**
UTILITY OF ANURAN CALL-SURVEY DATA FOR ESTIMATING OCCUPANCY, ABUNDANCE, AND REPRODUCTIVE SUCCESS IN THE CACHE RIVER BASIN

NEED: In Illinois eight of 41 amphibian species are listed as threatened or endangered and an additional six species have been identified as conservation priorities. Three of these species, the bird-voiced treefrog (*Hyla avivoca*), the crayfish frog (*Rana areolata*) and the wood frog (*Rana sylvatica*) have been identified during anuran call surveys (ACS) conducted in the Cache River basin of southern Illinois over the past five years. However, the use of ACS data to infer trends in occupancy and abundance of anuran species has been questioned (Bridges and Dorcas 2000; Crouch and Paton 2002; Oseen and Wassersug 2002; Nelson and Graves 2004; Weir et al. 2005). The presence of non-calling satellite males, for example, may cause these surveys to underestimate frog populations. The absence of calling frogs may be an indicator of environmental conditions during the survey period as well as frog abundance. Both of these factors could lead to estimators of occupancy and abundance that underestimate the true parameters. The presence of calling frogs, furthermore, does not necessarily indicate successful breeding or recruitment. For example, a wetland site may have breeding adults and even reproduction (egg laying), but it may dry too quickly (i.e. have a short hydroperiod) for the larvae to complete metamorphosis or it may contain predatory fish that prey on anuran larvae and prevent all metamorphosis. From a landscape perspective, this site would be considered a population sink, drawing migrants from nearby sites, some of which may be productive as a result of longer hydroperiods or absence of fish, thus wasting all reproductive effort. This would lead to estimators of occupancy and abundance that overestimate the true parameters.

In addition, when no frogs are detected at a wetland site, it may be the result of unsuitable habitat or it may be that it is isolated from other wetlands. In the latter case, some temporary disturbance may have occurred, such as introduction of fish, but the distance to the nearest wetland is too great to allow re-colonization. Although frogs have traditionally been considered organisms with limited dispersal capabilities, this view has been increasingly brought into question as studies have shown that anuran populations are greatly affected by the movement of individuals between habitats (Marsh and Trenham 2000). The need exists to determine effective colonization distances for anurans.

The need exists to critically evaluate the results of ACS so that any trends that are inferred from these data are accurate, as management decisions may be based on call-survey results. The evaluation of ACS data can be accomplished by comparing the results of in-depth field surveys with the results of call-survey data. The results from such a comparison can be extrapolated to call-survey data collected throughout Illinois. The need also exists to understand why some sites are sinks. This could result in management recommendations that would restore sinks into sources.

OBJECTIVES:

Job 1. Analyze existing frog calling survey data.

Examine attributes of habitat preferred by the three frog species at the ten Cache River wetland sites where volunteers have been conducting call-surveys for the past five years. Time frame: 15 April 2007 to 1 February 2008. Estimated Cost: $5,000.

Job 2. Estimate occupancy, abundance, reproductive success, and fine-scale habitat variables at the ten Cache River wetland sites where volunteers have been conducting call-surveys for the past five years.

2.1. Examine frog populations via field surveys at the ten sites. Time frame: 15 April 2007 to 30 August 2009. Estimated Cost: $35,000.
2.2. Measure fine-scale habitat attributes at the ten sites. Time frame: 15 April 2007 to 30 June 2009. Estimated Cost: $28,000.
2.3. Conduct regression analyses. Time frame: 1 July 2009 to 30 December 2009. Estimated Cost: $4,000.

Job 3. Conduct overall multiple regression analysis on the data collected in Jobs 1 and 2. Time Frame: 1 November 2009 to 1 February 2010. Estimated Cost: $3,000.

Job 4. Prepare annual performance reports to the Federal Aid Coordinator. Time frame: Annually. Estimated Cost: $2,000.

Job 5. Prepare final report. Time frame: 30 November 2009 to 30 March 2010. Estimated Cost: $3,000.

ACCOMPLISHMENTS

Job 1

Anuran call surveys have been conducted in the Cache River Basin since 1995. Some of the routes established at that time are still in use today, but others have been established more recently and have only been surveyed for one or two years. We selected Routes 1, 2, 4, 5, 6, and 7 for this project based on completeness of data. Geographic information system (GIS; ArcGIS V9.2, Environmental Systems Research Institute, Redlands, CA) was used to map the locations of the call sights (Figure 1). Geospatial data from the USGS National Land Cover Dataset were used to estimate the relative areas of agriculture, wetland, grassland, forest, and developed area within 500m and 1000m buffer zones of each call site. The crops (soybeans, pasture/hay, corn, double-cropped soybeans & winter wheat, rice, sorghum, and alfalfa) in each agriculture area were recorded. Wetland areas included both wetlands and open water areas. Developed areas ranged from developed open spaces to high intensity development. Road and stream density (sum of lengths) were also measured for each site at both buffer distances using data from the Illinois Natural Resources Geospatial Data Clearinghouse.

Job 2.1

We reviewed the pre-existing ACS data from the Cache River basin in southern Illinois and evaluated the completeness of the data for each route and the total length of time that each route had been surveyed. Based on this, we selected Route 2, which included ten sites located in the Cache River State Natural Area as shown in Figure 2. Between March 2007 and June 2008, these field sites in the Cache River State Natural Area were surveyed weekly for frog occupancy. This was done using a variety of methods, including visual, call, minnow trap, and dip net surveys. Visual surveys consisted of a 10 minute daytime sweep through each site. An observer would visually search for any evidence of frogs, including eggs, tadpoles, metamorphosed individuals, and adults, and identified any detected anurans to species. Call surveys always began shortly after sunset and ended before 2am according to the NAAMP protocol. During each call survey, an observer listened intently at a given site for 5 minutes, identifying any species of frog calling and rating the strength of the call chorus based on the calling index created by NAAMP. For each minnow trap survey, five collapsible minnow traps were set at each site overnight. The following day the contents of the trap were inspected, any frogs identified and released. The dip net survey at a site consisted of five sweeps with a dip net through the wetland, each sweep lasting 10 seconds. At the end of each sweep the contents of the net was inspected and any captured frogs or tadpoles identified. If, during the use of any of these survey methods, an individual was captured that could not be identified in the field, especially egg or larval specimens, it was collected and raised in captivity until the individual's identity could be confirmed.

The results of these surveys are shown in Table 1. For the purpose of this study it is assumed that the presence of metamorphs indicates survival to adulthood and successful recruitment. No evidence for reproduction was found for *Bufo americanus* or *Rana areolata* at any of the sites where they were detected with call surveys. Given the above assumption concerning the presence of metamorphs, evidence for recruitment was found for seven species: *Acris crepitans, Bufo fowleri, Hyla chrysoscelis, Pseudacris crucifer, P. triseriata, Rana catesbeiana, R. clamitans,* and *R. sphenocephala.*

Job 2.2
During each survey at each site fine-scale habitat attributes were measured and recorded.

Job 2.3
The presence or absence of a species was noted based on past ACS data for each calling site (Table 2). Multiple linear regression was used to evaluate the influence of the habitat attributes within buffers of 500 and 1000m on the presence or absence of a given species for all of the call survey sights located within the Cache River Basin (Table 3). The Hosmer and Lemeshow Goodness-of-Fit Test was used to assess the validity of each model (Table 4). These tests indicated that four models had questionable fits, *Hyla cinerea* at 500m, *Pseudacris streckeri* at 500m, *Rana clamitans* at 1000m, and *Rana palustris* at 1000m with p-values of 0.0202, 0.0197, 0.0031, and 0.0044. Since these were the only models with questionable fits and since, for each species, the model for one, but not both buffer zones was questionable, these were regarded as statistical artifacts and the models were kept for the analyses. Outliers were assessed using Cook's D and the diagonal elements of the hat matrix for each dataset. If the outlying sites had

no significant effect on the inferences made or if they had a significant effect as a result of being one of the very few (less than 5) sites where the species was detected (for rare species) or not detected (for common species) then the sites were not removed from the analysis. A total of two outlying sites were removed from the 500m buffer and one from the 1000m buffer analyses of *P. triseriata*. No other species had outliers removed from their analyses.

For three models the logistic regression convergence criterion were not met. For the 1000m *Acris crepitans* model, complete separation of data points was found and for both the 500m and 1000m *Pseudacris triseriata* models there was quasi-complete separation of data points. The convergence criterion for *A. creptians* at 1000m are satisfied if developed, agricultural, forested, or wetland area or road length is removed from the model. This means that each of these variables is a perfect predictor of the presence or absence of *A. crepitans* at a site when a buffer of 1000m is used and the other variables are included in the model. For both *P. triseriata* models, the removal of developed, agricultural, or forested areas from the model resulted in the satisfaction of the convergence criterion. The 500m *P. triseriata* model also converged if wetland area was removed and the 1000m model was additionally satisfied with the removal of either road or stream length. Each of these problem variables is a nearly perfect predictor of *P. triseriata* presence/absence at the respective buffer zone when the other variables are included in the model. The presence of complete or quasi-complete separation of data points in these three models is most likely an artifact of the low numbers of sites for which each species was absent; two sites for *A. crepitans* and 3 sites for *P. triseriata*. Because of these convergence problems, however, conclusions made with these three models are suspect.

The results from the multiple linear regression are shown in Table 3. Very few species showed significant ($p\text{-value} < 0.05$) effects from the habitat variables at either of the buffer zone distances. *Bufo. americanus* had a significant effect from wetland area within both buffer zone distances and additional significant effects from agriculture and forested areas within the 1000m buffer zones. It should be noted that for *B. americanus*, both agriculture and forested areas were close to significance within the 500m buffer zones with p-values of 0.0715 and 0.0591 respectively. Forested area had significant effects on the presence/absence of *H. cinerea* within both buffer zones, but no other habitat variables appear to be contributing the presence/absence of this species. The presence/absence of *P. streckeri* appears to be significantly affected only by developed, agricultural, forested, and wetland areas within the 1000m buffer, but within the 500m buffer all four of these variables had p-values of 0.0928, 0.0634, 0.0625, and 0.0569 respectively. *Rana. areolata* was significantly affected by wetland area within both buffer areas but by stream length only with the 500m buffer zone. Similarly, the presence/absence of *R. catesbeiana* was significantly affected by forested areas and road length within both buffer zones, but by developed area only with the 500m buffer zone. Wetland area and road length both had significant impacts on the presence/absence of *R. clamitans* within the 1000m buffer zone, but were only close to significance within the 500m area, p-values of 0.0930 and 0.0664 respectively.

There appears to be no pattern within genera for the impact of habitat variables on frog species. All the genera within which significant effects were found had only one member affected with the exception of *Rana*. *Rana* is also the largest genus in this study and is therefore, by chance, the most likely to have more than one species significantly affected by habitat variables. It is interesting to note that for those species significantly affected by the habitat variables, the number of sites where they were present is relatively equal to the number of sites where they were recorded absent. This is in contrast to those species whose presence/absence was not influenced by the habitat variables and were more often found at almost all or almost none of the sample sites. It is likely that a larger number of sites is needed to clarify the effects of habitat variables on the presence or absence of frog species.

Although care was taken when choosing the buffer sizes in this study, it is possible that the use of a larger scale analysis could change the end results. Houlahan and Findlay (2003) found that a larger landscape scale between 2000 and 4000 m is critical when examining species richness. They argued that the distance at which habitat variables become significant for a species is representative of that species' life history and that it should be expected that not all species will have habitat variables significant at the same scale. Since similar results were also found by Findlay and Houlahan (1997) and Hecnar and M'Closkey (1998), it is possible that expanding the buffer size and adding additional buffer zones in this study would result in more habitat variables influencing the presence/absence of individual anuran species. It is also possible that appropriate buffer size differs between species. Further work is needed to examine these issues.

In this study, wetland area was a significant influence in the presence/absence of *B. americanus, P. streckeri, R. areolata,* and *R. clamitans*. Weir et al. (2005) found that the proportion of palustrine wetlands within a 1km zone of a survey location explained variation in presence/absence for nearly all of the anuran species in their study and, similarly, Trenham et al. (2003) found that the number of wetland patches in a landscape was significantly associated with the detection of *P. triseriata, P. crucifer,* and *H. chrysoscelis* at a site. Considering the importance of wetland habitat in the life cycle of most frog species, its influence in this study was expected.

The introduction of roads into a landscape can create barriers to dispersal, habitat fragmentation, and road-related mortality. Eigenbrod et al. (2008) asserted that the negative impact of roads is largely due to road-related mortalities. They argued that the length of road in a landscape is less important than the amount of traffic on those roads. In this study only two species, *R. catesbeiana* and *R. clamitans,* were found to be significantly impacted by road length within the buffer zones. It is not unexpected that so few species showed significance for road length; most of our sites are located in a very rural area and the amount of traffic these roads are expected to receive should be minimal. Other studies have found strong associations between the frequency of road occurrence and the presence of frog species, but care needs to be taken to account for differences in traffic flow between study areas (Houlahan and Findlay 2003, Trenham et al. 2003).

Developed area in a landscape causes many of the same problems as, and in many cases is correlated with, road density. In this study, the presence of both *R. catesbeiana* and *P. streckeri* were found to be significantly influenced by the amount of developed area. Other studies have found negative associations between urban areas and many anuran species (Knutson et al. 1999, Pellet et al. 2004). Knutson et al. (1999) noted that urban areas are unsuitable for most frog species because of habitat loss and fragmentation, wetland contamination from runoff, and the stocking of urban wetlands with fish.

Forested area had a significant impact on *B. americanus, H. cinerea, P. streckeri,* and *R. catesbeiana*. Similar results linking frog presence with forested areas have been found in other studies (Knutson et al. 1999, Guerry and Hunter 2002, Houlahan and Findlay 2003, Trenham et al. 2003). It is surprising so few of the frog species in this study showed a significant relationship with forested areas since this habitat plays an important role in the life cycles of so many species. It is possible that there was not enough variation in the amount of forest at these rural sites to allow for statistical significance.

Knutson et al. (1999) found that the length of small streams in a landscape did not greatly influence the presence/absence of frog species. This supports the lack of significance of stream length observed in this study; only the presence of *R. areolata* was impacted to any extent by stream length within a 500m but not a 1000m radius. This lack of impact of stream length is not surprising. Most amphibian species rely on ephemeral wetlands to breed as these wetlands, unlike the more permanent streams, cannot support long term populations of predatory fish populations. Additionally, the larvae of most amphibian species are relatively weak swimmers and so the current imposed on them by a stream environment would negatively impact their survival.

In this study, agriculture was found to be strongly associated with the presence of only two species, *B. americanus* and *P. streckeri.* Trenham et al. (2003) found a strong negative association between the presence of *H. chrysoscelis* and cultivation, but a positive association for *P. crucifer* and cultivation. Conflicting results were also discussed by Knutson et al. (1999). In their study, a positive association was found between agricultural area and anurans in Wisconsin but not in Iowa. It is expected that agriculture would have a negative impact on anuran species as it causes a loss in suitable habitat, fragmentation in the landscape, and the introduction of harmful pesticides and herbicides into nearby aquatic environments, but it is possible that other landscape variables, such as forested areas may mitigate the effects of agriculture (Knutson et al. 1999). Further work needs to be done to clarify these relationships.

Job 3

To combine the fine-scale habitat and landscape variables into one analysis, the effectiveness of ACS data for predicting each species at each of the sites along Route 2 was determined. The presence of eggs, tadpoles, or metamorphs of a species was considered confirmation of that species having bred and the presence of metamorphs was considered evidence of that species having survived to adulthood at that site. A summary of the presence of calling individuals and detection of breeding and survival of each species is presented in Table 5.

An ANOVA of the number of levels of effectiveness of ACS data, i.e. ACS detection but not physically detected, ACS detection and reproduction detected, etc., for each species was conducted. The counts of each comparison category for each species satisfies the assumption of independence but the small sample sizes in this study are likely obscuring a clear picture of normality we would expect to see. A second consequence of the small sample sizes in this study is that no observations were removed as outliers from the analysis.

Tables 1 and 5 indicate that call surveys seem to be correlated with the breeding and survival to adulthood of some species but not all. Every species had at least one site where it was only detected by call surveys and several species were only detected by call surveys. In some species evidence for breeding was sometimes found, without evidence of survival to adulthood. In four instances, furthermore, individuals were detected via the physical survey methods but not by the call surveys.

Table 6 is an ANOVA table showing a significant difference in the number of instances for each species that physical evidence for breeding and/or survival were predicted by ACS data. Figure 3 shows the differences in results for each species.

The results of our study suggest that in most cases ACS data overestimate the reproductive and recruitment success in anuran species. This is a critical finding for conservation and management programs that based their strategies and efforts on ACS data. We suggest that ACS data should be used in conjunction with physical surveys to accurately represent healthy, self-sustaining anuran populations. In this study the sample size was too small to evaluate species differences in the predictive ability of call surveys. Figure 3 however, suggests that differences may be present between species based on the differences in the comparisons between ACS and physical data. We would expect there to be a similar comparison between ACS and physical data for all species at each site if species is not a factor affecting whether breeding and/or survival were detected. This is not the case, however, so ACS data may be differentially effective at predicting reproduction and recruitment success between species.

Variation in detection probabilities are also a problem in our study. Weir et al. (2005) note that in anuran call surveys, environmental conditions such as wind, air temperature, moonlight, observer skill, and noise disturbance in the area may affect the detection probabilities. Detection probabilities are an issue for each of the physical surveys as a result of differing environmental conditions and the physical characteristics of each site such as the amount of aquatic vegetation and water turbidity. Detection probabilities may also differ based on the physical and behavioral characteristics of each species. For example, those species which have larger, more visible egg masses and tadpoles would be expected to have a higher probability of detection. From Figure 3, however, there does not appear to be any readily discernable pattern among species with similar characteristics or among genera, but it is possible that the small sample sizes are obscuring any patterns that may exist.

<u>Job 4</u>
The annual and interim reports were prepared and distributed to agency staff.

<u>Job 5</u>
The final report was prepared and distributed to agency staff.

<u>Literature Cited</u>

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Tables and Figures

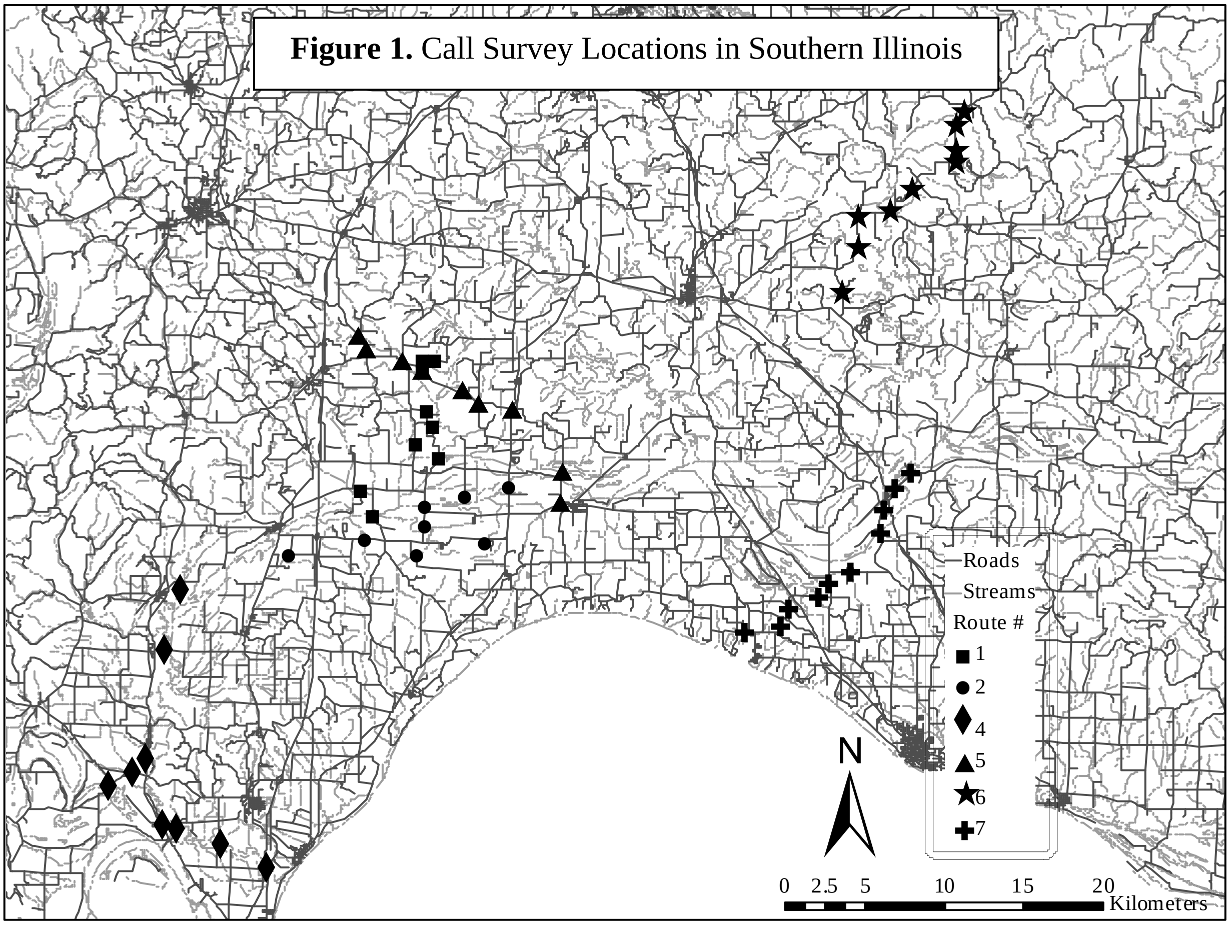

Figure 1. Call Survey Locations in Southern Illinois

Table 1. Survey Results Summary for Each Site

Site	Species	Maximum Calling Index	Adult	Egg Mass	Tadpole	Tad. w/ legs	Metamorph
1	*Acris crepitans*	3	0	0	0	0	0
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	1	0	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	1	0	0	0	0	0
	Pseudacris crucifer	3	6	0	0	0	0
	Pseudacris triseriata	1	0	0	0	0	0
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	2	8	0	0	0	0
	Rana clamitans	0	3	0	0	0	0
	Rana sphenocephala	0	29	0	0	0	0
2a	*Acris crepitans*	3	0	0	0	0	0
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	1	0	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	0	0	0	0	0	0
	Hyla chrysoscelis	1	0	0	0	0	0
	Pseudacris crucifer	1	0	0	0	0	0
	Pseudacris triseriata	0	0	0	0	0	0
	Rana aureolata	1	0	0	0	0	0
	Rana catesbeiana	1	1	0	0	0	0
	Rana clamitans	1	4	0	0	0	0
	Rana sphenocephala	1	5	0	0	0	0
2b	*Acris crepitans*	3	3	0	0	0	0
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	0	0	0	0	0	0
	Hyla avivoca	3	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	0	0	0	0	0	0
	Pseudacris crucifer	2	0	0	0	0	0
	Pseudacris triseriata	0	0	0	0	0	0
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	1	3	0	0	0	0
	Rana clamitans	1	0	0	0	0	0
	Rana sphenocephala	3	0	0	0	0	1
3	*Acris crepitans*	3	27	0	0	0	13
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	1	0	0	0	0	0
	Hyla avivoca	2	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	1	0	0	0	5	0
	Pseudacris crucifer	3	0	0	0	0	0
	Pseudacris triseriata	0	0	0	0	0	0
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	1	21	0	13	0	0
	Rana clamitans	0	0	0	5	0	0
	Rana sphenocephala	2	15	0	48	6	9
4	*Acris crepitans*	3	14	0	0	0	29
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	0	0	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	0	0	1	0	0
	Hyla chrysoscelis	1	0	0	0	5	0
	Pseudacris crucifer	2	0	0	0	0	0
	Pseudacris triseriata	1	0	0	0	0	0
	Rana aureolata	1	0	0	0	0	0
	Rana catesbeiana	2	9	0	0	0	0
	Rana clamitans	1	0	0	0	0	0
	Rana sphenocephala	2	1	0	20	5	2

Table 1. (cont.)							
Site	**Species**	**Maximum Calling Index**	**Adult**	**Egg Mass**	**Tadpole**	**Tad. w/ legs**	**Metamorph**
5	*Acris crepitans*	3	44	0	0	0	3
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	2	1	0	0	0	2
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	2	0	0	0	5	0
	Pseudacris crucifer	3	0	0	0	0	0
	Pseudacris triseriata	1	0	0	0	0	0
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	1	4	0	0	5	2
	Rana clamitans	1	0	1	0	0	0
	Rana sphenocephala	2	13	25	166	41	0
6	*Acris crepitans*	2	1	0	0	0	0
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	1	4	0	0	0	5
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	2	0	0	0	0	0
	Pseudacris crucifer	3	0	0	0	0	0
	Pseudacris triseriata	3	3	17	23	0	0
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	1	1	0	0	0	3
	Rana clamitans	0	0	0	0	0	0
	Rana sphenocephala	1	28	64	418	964	159
7	*Acris crepitans*	3	4	0	0	0	0
	Bufo americanus	1	0	0	0	0	0
	Bufo fowleri	0	1	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	0	0	0	0	0
	Hyla chrysoscelis	3	1	28	10	0	0
	Pseudacris crucifer	3	0	0	0	6	1
	Pseudacris triseriata	3	6	25	34	19	26
	Rana aureolata	0	0	0	0	0	0
	Rana catesbeiana	2	57	0	285	11	18
	Rana clamitans	1	0	0	6	0	4
	Rana sphenocephala	2	5	3	19	8	7
8	*Acris crepitans*	3	26	0	0	0	5
	Bufo americanus	0	0	0	0	0	0
	Bufo fowleri	3	0	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	3	1	0	0	0	0
	Hyla chrysoscelis	2	0	0	0	0	1
	Pseudacris crucifer	2	0	0	6	6	0
	Pseudacris triseriata	1	0	0	0	0	0
	Rana aureolata	1	0	0	0	0	0
	Rana catesbeiana	2	2	0	5	0	0
	Rana clamitans	1	0	0	0	0	0
	Rana sphenocephala	2	0	0	5	20	0
10	*Acris crepitans*	0	5	0	0	0	0
	Bufo americanus	1	0	0	0	0	0
	Bufo fowleri	0	0	0	0	0	0
	Hyla avivoca	0	0	0	0	0	0
	Hyla cinerea	2	0	0	0	0	0
	Hyla chrysoscelis	2	0	0	0	0	0
	Pseudacris crucifer	2	0	0	0	0	0
	Pseudacris triseriata	3	0	2	0	0	0
	Rana aureolata	1	0	0	0	0	0
	Rana catesbeiana	1	4	0	0	0	0
	Rana clamitans	0	0	0	0	0	0
	Rana sphenocephala	2	2	24	2988	397	8

Table 2. Presence/Absent Sample Sizes for Each Species
*The sample sizes used in the analysis for *P. triseriata* differed from those listed here due to elimination of outliers. For modeling with a 500m buffer, *P. triseriata* had samples sizes of 52 and 3 and with a 1000m buffer, sizes of 51 and 3 for presence and absence respectively.

Species	No. of Sites Present	No. of Sites Absent
Acris crepitans	54	2
Bufo americanus	38	18
Bufo fowleri	52	4
Hyla avivoca	19	37
Hyla cinerea	34	22
Hyla chrysoscelis	51	5
Pseudacris crucifer	53	3
Pseudacris streckeri	25	31
*Pseudacris triseriata**	53	3
Rana aureolata	36	20
Rana blairi	14	42
Rana catesbeiana	39	17
Rana clamitans	31	25
Rana palustris	16	40
Rana sphenocephala	46	10
Rana sylvatica	22	34
Scaphiopus holbrookii	10	46

Table 3. Significance of Habitat Variables at Different Buffer Zones for Presence/Absence of Species

Species	Buffer	Source	Type III Estimate	Standard Error	Wald X^2	df	p-value
Acris crepitans	500m	Intercept	-4.6770	10.0189	0.2179	1	0.6409
		Developed Area	39.9987	27.9692	2.0452	1	0.1527
		Agriculture Area	6.1975	10.8482	0.3264	1	0.5678
		Forested Area	6.2626	10.2320	0.3746	1	0.5405
		Wetland Area	58.0028	62.0634	0.8734	1	0.3500
		Stream Length	-0.00012	0.00183	0.0043	1	0.9478
		Road Length	-0.00130	0.00217	0.3605	1	0.5482
	1000m	Intercept	-395.8	335.8	1.3890	1	0.2386
		Developed Area	1160.3	1027.8	1.2746	1	0.2589
		Agriculture Area	414.5	347.7	1.4214	1	0.2332
		Forested Area	373.3	314.7	1.4067	1	0.2356
		Wetland Area	964.1	773.6	1.5530	1	0.2127
		Stream Length	-0.0117	0.00992	1.3991	1	0.2369
		Road Length	0.00286	0.00377	0.5750	1	0.4483
Bufo americanus	500m	Intercept	24.2509	12.0108	4.0767	1	0.0435
		Developed Area	-18.8119	13.7652	1.8677	1	0.1717
		Agriculture Area	-21.9618	12.1869	3.2475	1	0.0715
		Forested Area	-23.0505	12.2139	3.5617	1	0.0591
		Wetland Area	-23.6051	12.1674	3.7637	1	0.0524
		Stream Length	-0.00013	0.000474	0.0783	1	0.7797
		Road Length	-0.00123	0.000748	2.7065	1	0.0999
	1000m	Intercept	22.7461	9.2710	6.0195	1	0.0141
		Developed Area	-17.8086	13.5327	1.7318	1	0.1882
		Agriculture Area	-20.0296	9.5093	4.4366	1	0.0352
		Forested Area	-21.0620	9.2797	5.1515	1	0.0232
		Wetland Area	-22.3818	9.5754	5.4636	1	0.0194
		Stream Length	0.000105	0.000194	0.2948	1	0.5872
		Road Length	-0.00061	0.000352	2.9919	1	0.0837
Bufo fowleri	500m	Intercept	-4.1239	6.8592	0.3615	1	0.5477
		Developed Area	21.7854	16.2358	1.8005	1	0.1797
		Agriculture Area	7.9006	7.2846	1.1763	1	0.2781
		Forested Area	6.2245	6.9578	0.8003	1	0.3710
		Wetland Area	31.2452	21.8110	2.0522	1	0.1520
		Stream Length	-0.00072	0.00112	0.4141	1	0.5199
		Road Length	-0.00134	0.00147	0.8281	1	0.3628
	1000m	Intercept	-10.7872	17.2689	0.3902	1	0.5322
		Developed Area	25.0064	37.5771	0.4428	1	0.5058
		Agriculture Area	18.0008	18.9440	0.9029	1	0.3420
		Forested Area	14.0355	17.2310	0.6635	1	0.4153
		Wetland Area	55.0227	44.7428	1.5123	1	0.2188
		Stream Length	-0.00050	0.000475	1.1018	1	0.2939
		Road Length	-0.00057	0.000745	0.5878	1	0.4433
Hyla avivoca	500m	Intercept	-1.7293	2.3491	0.5419	1	0.4616
		Developed Area	1.5942	6.4425	0.0612	1	0.8046
		Agriculture Area	2.0698	2.5884	0.6394	1	0.4239
		Forested Area	0.5014	2.6592	0.0355	1	0.8504
		Wetland Area	1.0302	3.1318	0.1082	1	0.7422
		Stream Length	-0.00019	0.000437	0.1893	1	0.6635
		Road Length	-0.00006	0.000714	0.0066	1	0.9353
	1000m	Intercept	-1.2443	2.2058	0.3182	1	0.5727
		Developed Area	-16.0160	12.2994	1.6957	1	0.1929
		Agriculture Area	1.1035	2.7373	0.1625	1	0.6869
		Forested Area	-1.0307	2.7174	0.1439	1	0.7045
		Wetland Area	-0.2769	3.4024	0.0068	1	0.9345
		Stream Length	0.000159	0.000179	0.6896	1	0.3728
		Road Length	0.000278	0.000335	0.7944	1	0.4063

Table 3. (cont.)							
Species	**Buffer**	**Source**	**Type III Estimate**	**Standard Error**	**Wald X^2**	**df**	**p-value**
Hyla cinerea	500m	Intercept	-3.3894	4.3881	0.5966	1	0.4399
		Developed Area	4.0493	8.2291	0.2421	1	0.6227
		Agriculture Area	3.9369	4.5620	0.7447	1	0.3881
		Forested Area	1.4199	4.6220	0.0944	1	0.7587
		Wetland Area	32.7926	12.1110	7.3315	1	0.0068
		Stream Length	-0.00056	0.000658	0.7236	1	0.3950
		Road Length	-0.00023	0.000932	0.0603	1	0.8061
	1000m	Intercept	-1.9581	2.8464	0.4733	1	0.4915
		Developed Area	11.6356	12.9178	0.8113	1	0.3677
		Agriculture Area	4.5486	3.4264	1.7623	1	0.1843
		Forested Area	0.4255	3.5205	0.0146	1	0.9038
		Wetland Area	31.8221	10.3670	9.4222	1	0.0021
		Stream Length	-0.00008	0.000244	0.1165	1	0.7328
		Road Length	-0.00065	0.000433	2.2752	1	0.1315
Hyla chrysoscelis	500m	Intercept	14.9521	21.3982	0.4883	1	0.4847
		Developed Area	-9.2390	23.7449	0.1514	1	0.6972
		Agriculture Area	-11.3559	21.5753	0.2770	1	0.5987
		Forested Area	-13.6875	21.2178	0.4162	1	0.5189
		Wetland Area	0.8850	24.9434	0.0013	1	0.9717
		Stream Length	-0.00126	0.00104	1.4725	1	0.2249
		Road Length	-0.00031	0.00130	0.0583	1	0.8092
	1000m	Intercept	13.5240	19.3388	0.4891	1	0.4844
		Developed Area	-13.8199	31.0263	0.1984	1	0.6560
		Agriculture Area	-10.1545	19.9826	0.2582	1	0.6113
		Forested Area	-11.6514	19.2717	0.3655	1	0.5455
		Wetland Area	31.6230	43.1459	0.5372	1	0.4636
		Stream Length	-0.00035	0.000373	0.8699	1	0.3510
		Road Length	-0.00015	0.000602	0.0656	1	0.7979
Pseudacris crucifer	500m	Intercept	16.1629	18.4715	0.7657	1	0.3816
		Developed Area	-54.3060	33.9026	2.5658	1	0.1092
		Agriculture Area	-14.1480	18.7211	0.5711	1	0.4498
		Forested Area	-14.1623	19.5001	0.5275	1	0.4677
		Wetland Area	-20.1715	19.4425	1.0764	1	0.2995
		Stream Length	0.00377	0.00228	2.7266	1	0.0987
		Road Length	0.00334	0.00239	1.9437	1	0.1633
	1000m	Intercept	16.9128	14.1819	1.4222	1	0.2330
		Developed Area	-37.7481	23.7457	2.5271	1	0.1119
		Agriculture Area	-13.2444	13.4262	0.9731	1	0.3239
		Forested Area	-13.3386	13.9432	0.9151	1	0.3387
		Wetland Area	-16.2363	14.2037	1.3067	1	0.2530
		Stream Length	0.000706	0.000543	1.6905	1	0.1935
		Road Length	-0.00007	0.000674	0.0102	1	0.9197
Pseudacris streckeri	500m	Intercept	11.4735	6.0930	3.5458	1	0.0597
		Developed Area	-15.3574	9.1371	2.8250	1	0.0928
		Agriculture Area	-11.3823	6.1308	3.4469	1	0.0634
		Forested Area	-11.5652	6.2081	3.4705	1	0.0625
		Wetland Area	-12.3417	6.4808	3.6265	1	0.0569
		Stream Length	0.000544	0.000453	1.4386	1	0.2304
		Road Length	-0.00047	0.000729	0.4218	1	0.5161
	1000m	Intercept	16.8526	6.7227	6.2842	1	0.0122
		Developed Area	-27.3251	13.6447	4.0105	1	0.0452
		Agriculture Area	-17.3974	7.0445	6.0991	1	0.0135
		Forested Area	-17.9139	6.8227	6.8938	1	0.0086
		Wetland Area	-17.1726	7.2557	5.6016	1	0.0179
		Stream Length	0.000227	0.000181	1.5845	1	0.2081
		Road Length	-0.00008	0.000339	0.0501	1	0.8229

Table 3. (cont.)							
Species	**Buffer**	**Source**	**Type III Estimate**	**Standard Error**	**Wald X^2**	**df**	**p-value**
Pseudacris triseriata	500m	Intercept	642.0	608.7	1.1124	1	0.2916
		Developed Area	-702.7	668.8	1.1039	1	0.2934
		Agriculture Area	-634.1	603.4	1.1042	1	0.2933
		Forested Area	-640.0	607.4	1.1100	1	0.2921
		Wetland Area	-620.0	592.9	1.0936	1	0.2957
		Stream Length	-0.00110	0.00237	0.2158	1	0.6422
		Road Length	0.00218	0.00352	0.3834	1	0.5358
	1000m	Intercept	298.0	528.5	0.3179	1	0.5729
		Developed Area	-315.3	339.7	0.8614	1	0.3534
		Agriculture Area	-248.6	398.6	0.3889	1	0.5329
		Forested Area	-297.9	525.7	0.3212	1	0.5709
		Wetland Area	-144.9	259.4	0.3123	1	0.5763
		Stream Length	-0.00683	0.0208	0.1079	1	0.7426
		Road Length	0.00189	0.00263	0.5193	1	0.4711
Rana aureolata	500m	Intercept	-0.2417	1.8071	0.0179	1	0.8936
		Developed Area	4.0961	6.5307	0.3934	1	0.5305
		Agriculture Area	2.9308	2.2217	1.7402	1	0.1871
		Forested Area	-0.0718	2.1498	0.0011	1	0.9733
		Wetland Area	8.3993	4.0746	4.2493	1	0.0393
		Stream Length	-0.00120	0.000558	4.6322	1	0.0314
		Road Length	-0.00035	0.000768	0.2052	1	0.6505
	1000m	Intercept	-0.9638	2.0782	0.2151	1	0.6428
		Developed Area	-1.9125	11.1785	0.0293	1	0.8642
		Agriculture Area	3.5002	2.6416	1.7556	1	0.1852
		Forested Area	0.2639	2.4521	0.0116	1	0.9143
		Wetland Area	8.0582	4.2192	3.6477	1	0.0561
		Stream Length	-0.00032	0.000200	0.1135	1	0.1085
		Road Length	0.000109	0.000325	2.5766	1	0.7362
Rana blairi	500m	Intercept	-0.9591	1.9947	0.2312	1	0.6306
		Developed Area	-1.7618	7.1021	0.0615	1	0.8041
		Agriculture Area	-0.0746	2.3371	0.0010	1	0.9745
		Forested Area	0.0909	2.3641	0.0015	1	0.9693
		Wetland Area	1.9876	2.8494	0.4866	1	0.4855
		Stream Length	-0.00001	0.000465	0.0005	1	0.9825
		Road Length	-0.00017	0.000793	0.0476	1	0.8274
	1000m	Intercept	-1.7651	2.5338	0.4853	1	0.4860
		Developed Area	-7.9225	12.3979	0.4084	1	0.5228
		Agriculture Area	-0.1316	3.0641	0.0018	1	0.9657
		Forested Area	-0.5152	3.0150	0.0292	1	0.8643
		Wetland Area	3.2136	3.5415	0.8234	1	0.3642
		Stream Length	0.000025	0.000193	0.0175	1	0.8949
		Road Length	0.000242	0.000349	0.4790	1	0.4889
Rana catesbeiana	500m	Intercept	-0.7440	1.8833	0.1561	1	0.6928
		Developed Area	14.3690	7.2285	3.9515	1	0.0468
		Agriculture Area	3.9600	2.3107	2.9371	1	0.0866
		Forested Area	5.2020	2.4983	4.3356	1	0.0373
		Wetland Area	4.0501	2.9369	1.9018	1	0.1679
		Stream Length	-0.00034	0.000471	7.1873	1	0.4667
		Road Length	-0.00231	0.000862	0.5298	1	0.0073
	1000m	Intercept	-0.9860	2.0881	0.2230	1	0.6368
		Developed Area	18.3528	11.4910	2.5509	1	0.1102
		Agriculture Area	4.5788	2.7364	2.7999	1	0.0943
		Forested Area	6.1658	2.9131	4.4797	1	0.0343
		Wetland Area	3.3837	3.1883	1.1263	1	0.2886
		Stream Length	-0.00010	0.000192	0.2585	1	0.6111
		Road Length	-0.00086	0.000368	5.4528	1	0.0195

Table 3. (cont.)							
Species	**Buffer**	**Source**	**Type III Estimate**	**Standard Error**	**Wald X^2**	**df**	**p-value**
Rana clamitans	500m	Intercept	0.3028	1.8099	0.0280	1	0.8671
		Developed Area	5.8846	6.5491	0.8074	1	0.3689
		Agriculture Area	0.7248	2.1444	0.1142	1	0.7354
		Forested Area	1.9082	2.1813	0.7653	1	0.3817
		Wetland Area	5.6304	3.3516	2.8222	1	0.0930
		Stream Length	-0.00029	0.000476	0.3591	1	0.5490
		Road Length	-0.00143	0.000780	3.3705	1	0.0664
	1000m	Intercept	-0.3752	1.9175	0.0383	1	0.8449
		Developed Area	11.6392	10.8558	1.1495	1	0.2836
		Agriculture Area	2.8569	2.4957	1.3103	1	0.2523
		Forested Area	2.9223	2.3672	1.5240	1	0.2170
		Wetland Area	10.3427	4.6397	4.9693	1	0.0258
		Stream Length	-0.00024	0.000199	1.5034	1	0.2202
		Road Length	-0.00066	0.000328	4.0585	1	0.0439
Rana palustris	500m	Intercept	-9.3543	7.3163	1.6347	1	0.2011
		Developed Area	4.9573	9.8986	0.2508	1	0.6165
		Agriculture Area	8.1365	7.4900	1.1801	1	0.2773
		Forested Area	9.2199	7.5169	1.5044	1	0.2200
		Wetland Area	8.1716	7.6230	1.1491	1	0.2837
		Stream Length	-0.00002	0.000459	0.0027	1	0.9587
		Road Length	0.000412	0.000727	0.3206	1	0.5712
	1000m	Intercept	-6.1521	6.5075	0.8937	1	0.3445
		Developed Area	-9.2971	13.9479	0.4443	1	0.5051
		Agriculture Area	4.9441	6.9913	0.5001	1	0.4795
		Forested Area	6.0936	6.6759	0.8332	1	0.3614
		Wetland Area	5.0639	7.2771	0.4842	1	0.4865
		Stream Length	0.000056	0.000189	0.0870	1	0.7680
		Road Length	0.000252	0.000336	0.5594	1	0.4545
Rana sphenocephala	500m	Intercept	23.1023	16.5923	1.9386	1	0.1638
		Developed Area	-32.3249	20.1376	2.5767	1	0.1085
		Agriculture Area	-21.3559	16.9931	1.5794	1	0.2088
		Forested Area	-21.8368	16.7326	1.6930	1	0.1932
		Wetland Area	-4.2879	18.4799	0.0538	1	0.8165
		Stream Length	-0.00033	0.000681	0.0125	1	0.6325
		Road Length	0.000111	0.000988	0.2286	1	0.9108
	1000m	Intercept	23.1592	14.9658	2.3947	1	0.1217
		Developed Area	-34.1202	20.7946	2.6923	1	0.1008
		Agriculture Area	-19.1061	15.2582	1.5680	1	0.2105
		Forested Area	-20.4203	14.7976	1.9043	1	0.1676
		Wetland Area	7.6483	22.2008	0.1187	1	0.7305
		Stream Length	-0.00016	0.000271	0.7659	1	0.5565
		Road Length	-0.00043	0.000487	0.3457	1	0.3815
Rana sylvatica	500m	Intercept	-0.2418	1.7975	0.0181	1	0.8930
		Developed Area	3.1541	6.3137	0.2496	1	0.6174
		Agriculture Area	-0.2621	2.1096	0.0154	1	0.9011
		Forested Area	1.2444	2.1339	0.3400	1	0.5598
		Wetland Area	2.9868	2.7277	1.1990	1	0.2735
		Stream Length	-0.00061	0.000445	1.8731	1	0.1711
		Road Length	-0.00050	0.000733	0.4598	1	0.4977
	1000m	Intercept	-0.0194	1.8687	0.0001	1	0.9917
		Developed Area	-15.9758	12.5282	1.6261	1	0.2022
		Agriculture Area	-1.4789	2.4254	0.3718	1	0.5420
		Forested Area	-0.6448	2.2632	0.0812	1	0.7757
		Wetland Area	2.9232	3.2064	0.8312	1	0.3619
		Stream Length	-0.00005	0.000190	0.0639	1	0.8004
		Road Length	0.000366	0.000330	1.2301	1	0.2674

Table 3. (cont.)							
Species	**Buffer**	**Source**	**Type III Estimate**	**Standard Error**	**Wald X^2**	**df**	**p-value**
Scaphiopus holbrooki	500m	Intercept	-0.7813	1.9677	0.1577	1	0.6913
		Developed Area	-1.0053	7.5334	0.0178	1	0.8938
		Agriculture Area	-1.1541	2.4090	0.2295	1	0.6319
		Forested Area	-0.6568	2.3981	0.0750	1	0.7842
		Wetland Area	-1.6641	3.3386	0.2484	1	0.6182
		Stream Length	-0.00020	0.000554	0.1252	1	0.7235
		Road Length	0.000253	0.000879	0.0830	1	0.7733
	1000m	Intercept	-0.9995	2.1248	0.2213	1	0.6381
		Developed Area	-1.9095	13.5466	0.0199	1	0.8879
		Agriculture Area	1.6774	2.8227	0.3531	1	0.5523
		Forested Area	0.0639	2.6020	0.0006	1	0.9804
		Wetland Area	1.7573	3.6945	0.2262	1	0.6343
		Stream Length	-0.00044	0.000294	2.2414	1	0.1344
		Road Length	0.000018	0.000376	0.0023	1	0.9616

Table 4. Results of the Hosmer and Lemeshow Goodness-of-Fit Tests				
Model Information		**X^2**	**df**	**p-value**
Species	**Buffer**			
Acris crepitans	500m	1.1136	6	0.9809
	1000m	0.0257	2	0.9872
Bufo americanus	500m	2.5753	8	0.9581
	1000m	5.8351	7	0.5591
Bufo fowleri	500m	3.4167	7	0.8440
	1000m	3.1200	7	0.8737
Hyla avivoca	500m	10.2920	8	0.2451
	1000m	7.9810	8	0.4353
Hyla cinerea	500m	18.1433	8	0.0202
	1000m	5.3819	7	0.6135
Hyla chrysoscelis	500m	7.9158	8	0.4417
	1000m	2.1145	8	0.9773
Pseudacris crucifer	500m	2.5954	8	0.9571
	1000m	4.8267	8	0.7759
Pseudacris streckeri	500m	18.2159	8	0.0197
	1000m	7.0403	7	0.4247
Pseudacris triseriata	500m	0.1884	4	0.9958
	1000m	0.0511	1	0.8212
Rana areolata	500m	4.5923	8	0.8001
	1000m	10.2281	8	0.2494
Rana blairi	500m	10.3571	8	0.2409
	1000m	7.9179	8	0.4415
Rana catesbeiana	500m	3.4901	8	0.9000
	1000m	14.8675	8	0.0618
Rana clamitans	500m	8.8907	8	0.3516
	1000m	23.1900	8	0.0031
Rana palustris	500m	5.8656	8	0.6623
	1000m	20.6217	7	0.0044
Rana sphenocephala	500m	4.7749	7	0.6874
	1000m	3.4474	7	0.8408
Rana sylvatica	500m	9.6822	7	0.2073
	1000m	9.9188	8	0.2708
Scaphiopus holbrookii	500m	7.3775	8	0.4965
	1000m	2.4392	7	0.9316

Table 5. Comparison of Call Surveys Results with Physical Evidence of Reproduction and Survival to Adulthood. N = No evidence for species presence; C = Call surveys indicate the presence of a species; B = Call surveys indicate presence and physical evidence of breeding; S = Call surveys indicate presence and physical evidence of breeding and survival; P = Physical evidence for presence, but no individuals heard calling.												
Site	*Acris crepitans*	*Bufo americanus*	*Bufo fowleri*	*Hyla avivoca*	*Hyla cinerea*	*Hyla chrysoscelis*	*Pseudacris crucifer*	*Pseudacris triseriata*	*Rana aureolata*	*Rana catesbeiana*	*Rana clamitans*	*Rana sphenocephala*
1	C	N	C	N	C	C	B	C	N	C	P	P
2a	C	N	C	N	N	C	C	N	C	C	C	C
2b	C	N	N	C	C	N	C	N	N	C	C	S
3	S	N	C	C	C	B	C	N	N	B	P	S
4	S	N	N	N	B	B	C	C	C	C	C	S
5	S	N	S	N	C	B	C	C	N	S	C	S
6	C	N	S	C	C	C	C	B	N	S	N	S
7	C	C	C	N	C	B	S	S	N	S	S	S
8	S	N	C	N	C	S	B	C	C	B	C	B
10	P	C	N	N	C	C	C	B	C	C	C	S

Table 6. ANOVA Table					
Dependent Variable: Count					
Source	DF	Type III Sum of Squares	Mean Square	F-Value	P-value > F
Model	15	124.6666667	8.3111111	1.75	0.0762
Species	11	0.0000000	0.0000000	0.00	1.0000
Result	4	124.6666667	31.1666667	6.55	0.0003
Error	44	209.3333333	4.7575758		
Corrected Total	59	334.0000000			
R-square = 0.373253					

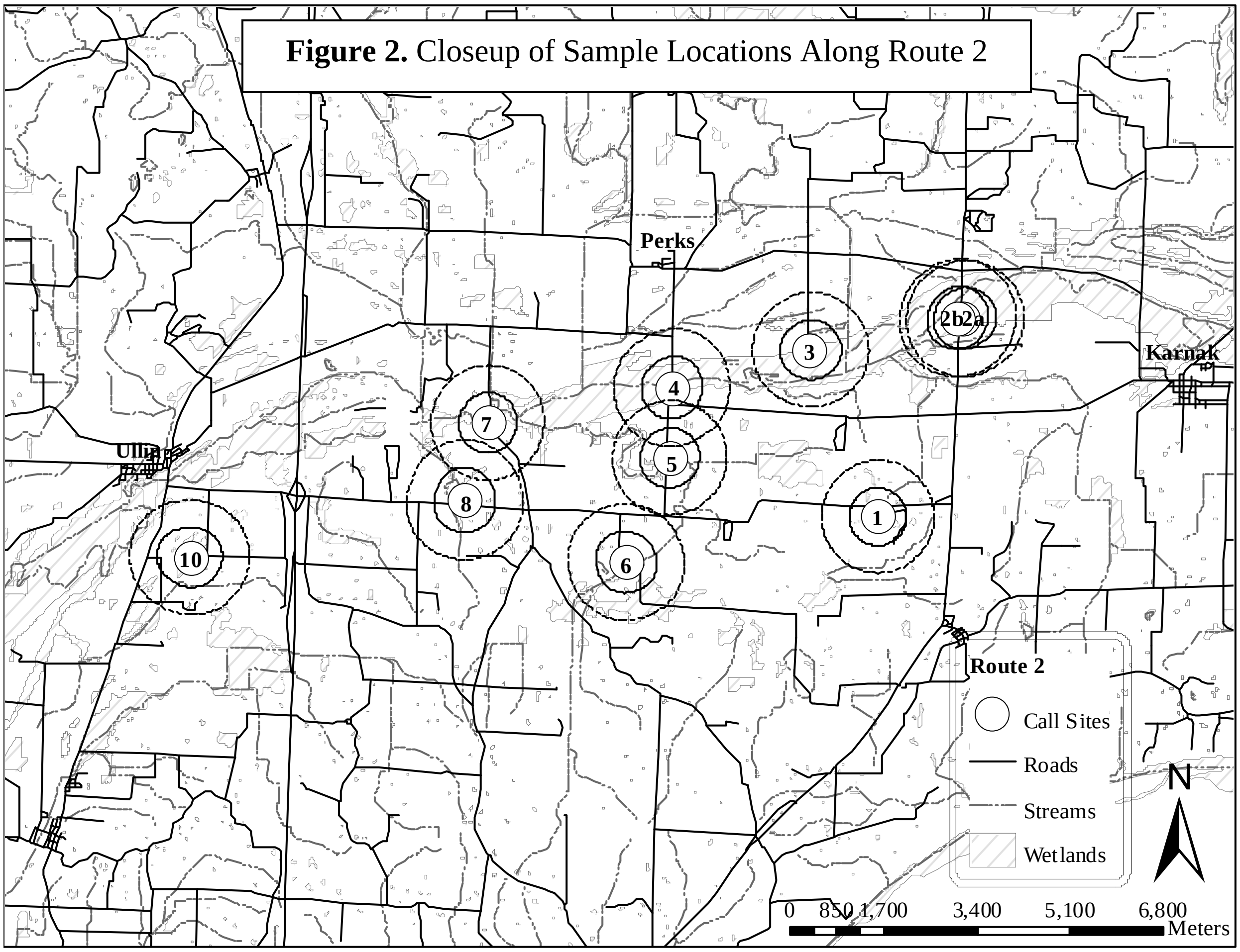

Figure 2. Closeup of Sample Locations Along Route 2

Figure 3. Comparison of the Predictive Ability of Call Surveys Among Species